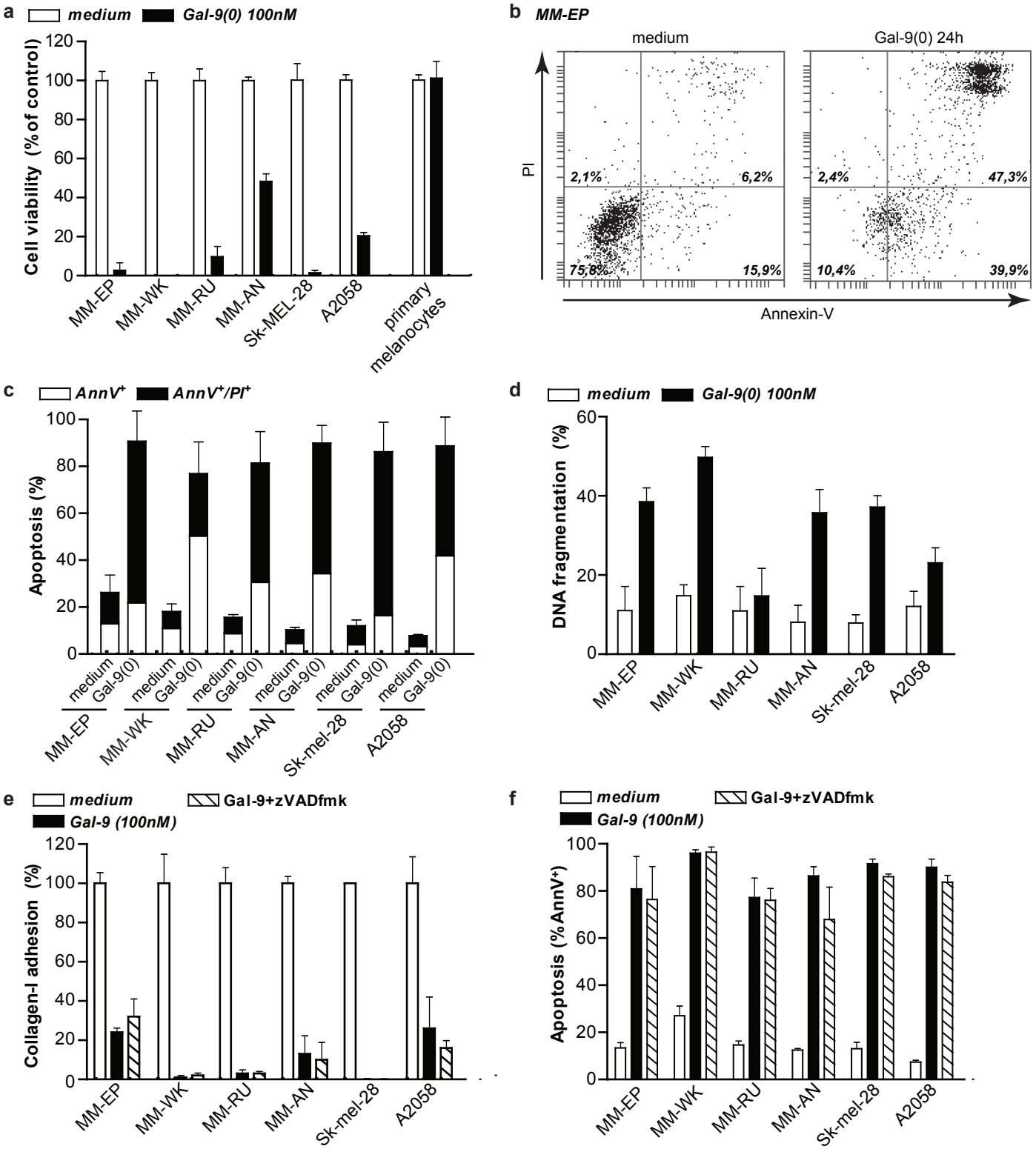
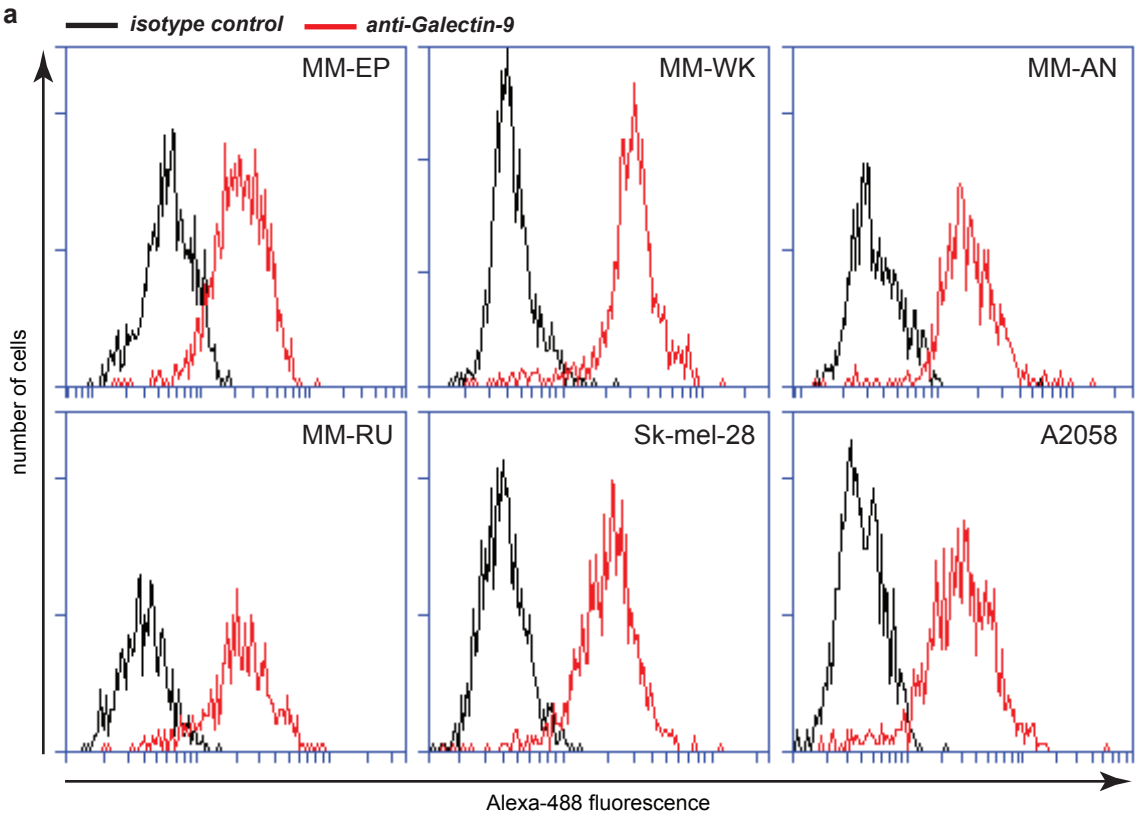
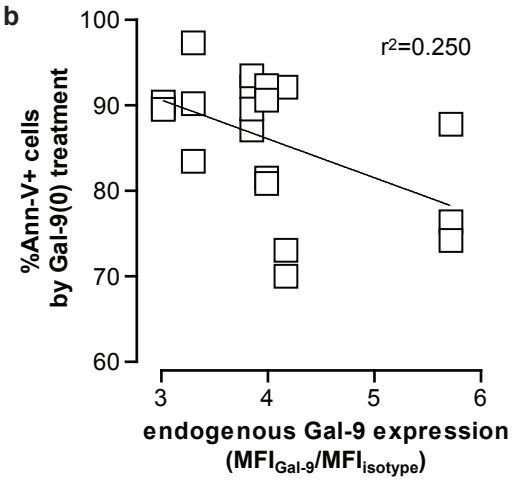
Supplementary Figure 1



Supplementary Figure 2





Legends to the Supplementary figures

Fig.S1. Gal-9(0) induces cell death in melanoma cells but not melanocytes. A. A panel of human melanoma cell and primary human melanocytes were treated with recombinant Gal-9 for 24h and analyzed for cell viability using MTS viability assay (Promega). In brief, $3x10^4$ cells/well were seeded into 48-well plates and allowed to adhere o.n.. Subsequently, cells were treated with 100nM Gal-9(0) for 24h. As a maximum cell death control, cells were treated for 20 minutes with 20% ethanol. Percentage viability was determined using the formula: $(OD450_{medium})-(OD450_{ethanol})/(OD450_{Gal-9})-OD450_{ethanol})x100\%$. Experiments were performed in triplicates. **B.** MM-EP cells were treated with 100nM Gal-9(0) for 24h and analyzed for cell death by Annexin-V/PI double staining. Dot-plots represent typical results obtained in medium control and Gal-9(0) treated cells. **C.** A panel of human melanoma cell lines was treated for 24h with 100nM Gal-9(0) and subsequently analyzed apoptosis by Annexin-V/PI staining. **D.** Cells were treated as in C, but were now analyzed by flow cytometry for DNA fragmentation using standard Propidium Iodide protocol. **E.** The inhibitory effect of Gal-9(0) on adhesion of human melanoma cell lines to Collagen-I coated wells is not blocked by pan-caspase inhibitor zVADfmk. In brief, $3x10^4$ melanoma cells were added to collagen-I coated plates and incubated for 1h at 37˚C with 100nM Gal-9(0) in the presence or absence of 40μM zVADfmk. Subsequently, wells were washed twice with phosphate buffered saline and the number of adhered cells was counted in three wells per condition. Experiments were performed in triplicates. **F.** Melanoma cell lines were treated with 100nM Gal-9(0) in the presence or absence of 40μM zVADfmk and analyzed for cell death by AnnexinV staining. All experimental values are mean + standard deviation of three independent experiments.

Fig.2. Expression of endogenous Galectin-9 in human melanoma cell lines. Representative histograms of a flow cytometric staining for endogenous Galectin-9 in a panel of human melanoma cell lines. In brief, 3x105 cells were fixed and permeabilized using standard protocol with FIX & PERM® reagent system (Life technologies) after which cells were stained using anti-Galectin-9 antibody FG-9 or IgG isotype control. Cells were washed twice with PBS and then stained using secondary Alexa-488 conjugated anti-mouse antibody. Cells were analyzed on an Accuri C6 flow cytometer (BD biosciences).